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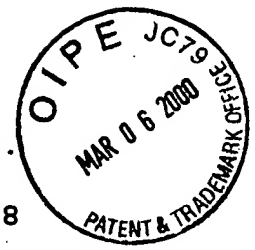
IN THE UNITED STATES PATENT AND TRADEMARK OFFICE



Attorney Docket No.:	**T5530.CIP**
Inventors:	**Rao et al.**
Serial No.:	**09/109,858**
Filing Date:	**July 2, 1998**
Examiner:	**J. Kerr**
Group Art Unit:	**1633**
Title:	**Lineage-restricted neuronal precursors**

Declaration by Dr. Mahendra Rao

I, Mahendra Rao, hereby declare that:

1. I am a co-inventor of the above-referenced patent application.

2. I have reviewed in detail the teachings of the reference by Blass-Kampmann et al. in Journal of Neuroscience Research 37:359-373 (1994).

3. The cells described by Blass-Kampmann et al. differ from our lineage-restricted neuronal precursors (NRP) cells which are described and claimed in the above-referenced patent application in their ability to differentiate into astrocytes, their growth factor requirements, and the time of isolation and/or survival.

4. Specifically, as taught at page 364-365 of this reference, only about half of the cells isolated differentiated into neurons. These cells also differentiated into glial cells

such as astrocytes. See page 366. In contrast, as demonstrated in experiments described in detail in Examples 3, 4 and 5 of the above-referenced patent application, no NRP cells of the present invention differentiated into astrocytes or oligodendrocytes even when the cells were grown under conditions known to promote astrocyte and oligodendrocyte differentiation in other neuroepithelial stem cells.

5. Further, at page 362, Blass-Kampmann teaches that they grew their cells in 10% FCS. As taught in the above-referenced patent application in Example 5, the NRP cells of the present invention die when grown in 10% FCS.

6. As also taught in the first paragraph at page 362 of Blass-Kampmann et al., their cells grew and survived in the absence of FGF. In contrast, the NRP cells of the present invention require FGF for survival and proliferation. See specifically, Example 3 of the above-referenced patent application.

7. Finally, as discussed at page 364 of Blass-Kampmann et al. in the last paragraph, their cells died after three days in culture. However, as discussed in Example 7 of the above-referenced patent application, the NRP cells of the present invention grow over multiple passages in mass and clonal culture.

8. I have also reviewed in detail the teachings of Boss et al. (U.S. Patent 5,411,883).

9. The cells described by Boss et al. also differ from our NRP cells in their ability to differentiate into different neurons and their growth factor requirements.

10. Specifically, Boss et al. describe a mesencephalic cell that differentiates into dopaminergic neurons. In contrast, the NRP cells of the present invention differentiate primarily into cholinergic and glutaminergic neurons.

11. Further, in col. 7, lines 55-60, Boss et al. show that growth in F12 with 5% serum is sufficient to maintain growing cells. In contrast, the NRP cells of the present invention require FGF for survival and proliferation. See specifically, Example 3 of the above-referenced patent application.

12. It appears that the Examiner may not have appreciated an important distinction between methods for obtaining neurons such as described by Weiss et al., Rao et al., Johe et al. and Lee et al. and obtaining dividing neuronal precursors. Neurons can be obtained from cell populations via methods such as those taught by Johe et al., Rao et al., Weiss et al. or Lee et al. However, dividing neuronal precursors that differentiate solely into neurons and not into glial cells had not been shown to

develop from any of the cell populations described in the art. Indeed controversy existed as to even the existence of such dividing neuronal restricted precursors until we presented unambiguous data to show that such a precursor existed and that this precursor had a characteristic set of properties. See for example Kalyani, A. and Rao, M.S. Cell Biol. Biochem. 1998 76:1-17 and Rao M.S. Anal. Record 1999 257:137-148 which are attached hereto as Exhibit A.

I further declare that all statements made herein of my own knowledge are true, and that all statements made on information and belief are believed to be true and further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Mahendra Rao

02/04/00

Date